



# PHOTOPHYSICAL STUDY OF ROSE BENGAL DYE IN AQUEOUS MICELLAR SOLUTION

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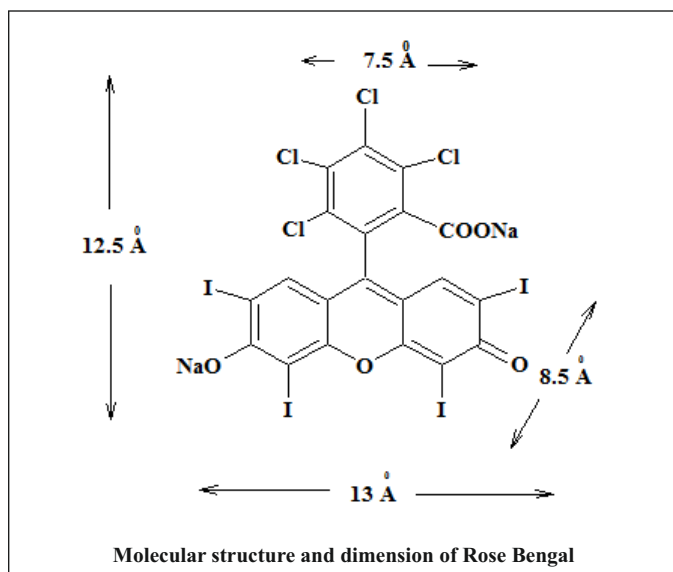
## ABSTRACT

The absorption and fluorescence spectra of Rose Bengal dye were studied in various solvents. It was found that solvent effects on the absorption wavelength are consistent with the solvatochromic shift. We found a linear relationship with the absorption wavelength of the dye in various solvents. Calculations were used to assign the absorption in the region 540–570nm to a  $\pi-\pi^*$  transition between the HOMO and LUMO of the anion. Experimental ground state and excited state dipole moments were calculated by using the solvatochromatic shifts of absorption and fluorescence spectra.

**KEY WORD:** Absorption, Fluorescence Spectra, Rose Bengal, Solvatochromic shift.

## 1. INTRODUCTION:

Rose Bengal (RB) (4,5,6,7-tetrachloro-2',4',5',7'-tetraiodofluorescein) is a tetraiodosubstituted dye of the xanthene class of dyes. It exhibits unusual spectroscopic and photochemical properties including a large absorption coefficient in the visible region and a high tendency for intersystem crossing to produce a photochemically active triplet excited state. The dye has been used in photodynamic inactivation of catalase, photoinactivation of olefins via the production of singlet oxygen, as a photosensitizing agent for inactivating biological species such as vaccinia virus, microsomal glucose-6-phosphatase, trypsin, Escherichia coli, acetylcholinesterase, and HL-60 cells [1-3]. Despite the numerous applications of Rose Bengal dye in various areas, information on the spectral properties of the dye in various solvents required for understanding its photochemical behavior seems to be incomplete.



Rose Bengal is a biological stain, its sodium salt is commonly used in eye drops to stain damaged conjunctival and corneal cells and thereby identify damage to the eye[4]. The stain is also used in the preparation of Foraminifera for microscopic analysis, allowing the distinction between forms that were alive or dead at the time of collection. A form of Rose Bengal is also being studied as a treatment for certain cancers and skin conditions[5]. The cancer formulation of the drug, known as PV-10, is currently undergoing clinical trials for melanoma and breast cancer. The company also has formulated a drug based on Rose Bengal for the treatment of eczema and psoriasis; this drug, PH-10, is currently in clinical trial as well. Rose Bengal is also used in synthetic chemistry to generate singlet oxygen from triplet oxygen. The singlet oxygen can then undergo a variety of useful reactions, particularly [2 + 2] cycloadditions with alkenes and similar systems.

Rose Bengal is used in animal models of ischemic stroke (photothrombotic stroke models) in biomedical research. A bolus of the compound is injected into the venous system. Then the region of interest, e. g. the cerebral cortex, is exposed and illuminated by LASER light of 561 nm. A thrombus is formed in the illuminated blood vessels, causing a stroke in the dependent brain tissue. Rose

Bengal is being researched as an agent in creating nano sutures.[6] Wounds are painted on both sides with it and then illuminated with an intense light. This links the tiny collagen fibers together sealing the wound.[7-9]

Rose Bengal, disodium salt, holds an important position among all dyes for a number of reasons [10]

1. It has a large absorption in all solvents in which it is soluble which corresponds almost exactly to the sodium D line.
2. Its intersystem crossing yield is high, but is not unity. All Rose Bengals show some fluorescence.
3. Its triplet is completely quenched by oxygen.
4. Its spectrum is most diagnostic of its immediate environment.
5. It bleaches slowly in protic, polar solvents.
6. It is a photodynamic sensitizer.
7. Its singlet may be quenched by strong oxidizing agents, in some cases forming radicals.

## 2. EXPERIMENTAL:

### 2.1 Chemicals:

Rose Bengal from Loba Chemia AR grade is used without additional purification. However spectra purity of compound was tested by taking their melting point and production of similar spectra when excited at different wavelengths. Sodium dodecyl sulphate (SDS), Cetyl tri methyl ammonium bromide (CTAB) Cationic surfactant, Brij -35 (non ionic surfactant) were purchased from SD Fine-Chem Ltd.

### 2.2. Preparation of solutions:

Predetermined amounts of the Rose Bengal dissolved in distilled water to obtained 1 mM stock solution. 1mM Triethyl amine,  $1 \times 10^{-2}$  M of SDS solution is used as stock solution.

## 3. RESULTS AND DISCUSSION:

Absorption and emission spectra of Rose Bengal were recorded in Water, Ethanol and Micellar solution of CTAB, SDS, Brij-35. The absorption spectra of RB shown in Fig. 1. are structure less, broad bands except in ethanol, CTAB and Water with a shoulder band in the region of 490nm to 530 nm. The values of absorption maxima are given in Table.1. Electronic spectrum for Rose Bengal anion in methanol shows a strong absorption (oscillator strength 0.46) at 503nm that arises primarily from a HOMO–LUMO transition of  $\pi \rightarrow \pi^*$  character.[11]

The dye solution shows a significant solvatochromic shift of about 700  $\text{cm}^{-1}$  from its aqueous water solution to non aqueous ethanol solution. However the absorption maxima are not much changed in micellar solution. The smaller shift implies that the ground state energy distribution in the dye is not effected due to the polar nature of the solvents. On the other hand a larger shift indicates specific interaction between the solute and solvents molecules. Large solvatochromic shifts are generally associated with charge transfer bands. The shifts in absorption energy for Rose Bengal are less than 800  $\text{cm}^{-1}$  and consistent with the assignment of the absorption to a  $\pi-\pi^*$  transition [12].

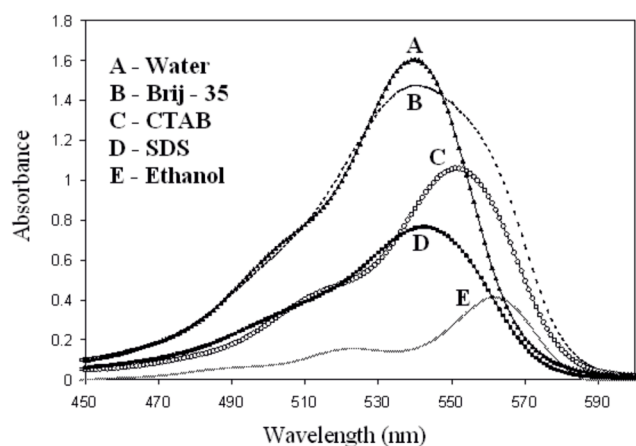


Fig. 3.1. Absorption spectrum of Rose Bengal at different media

This implies that the dipole moment of the dye molecule in the excited state is more than in the ground state [44].

The excitation and fluorescence spectra of solution of  $1 \times 10^{-5}$  M are shown in Fig. 2. and Fig.3. are also structureless broad bands. The values of  $\lambda_{em}$  and  $\lambda_{ex}$  are given in same Table. 1. The analysis of solvent effect on spectral properties of dye solutions was carried out by using the spectral shift in various solvents.

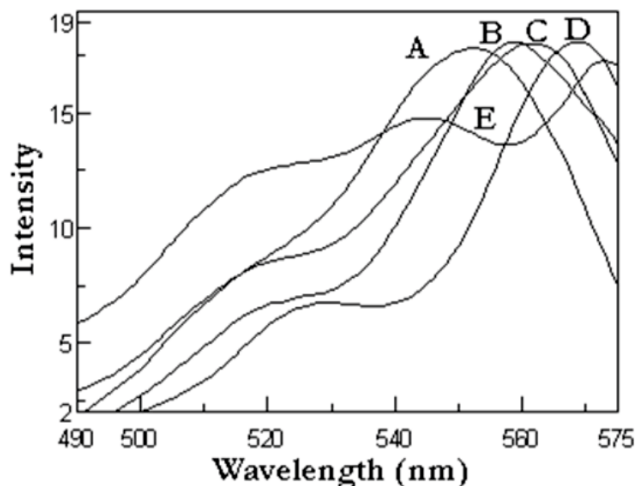


Fig. 2. Excitation spectra of RB in CTAB (A), Brij-35 (B), SDS (C), Ethanol (D), Water (E)

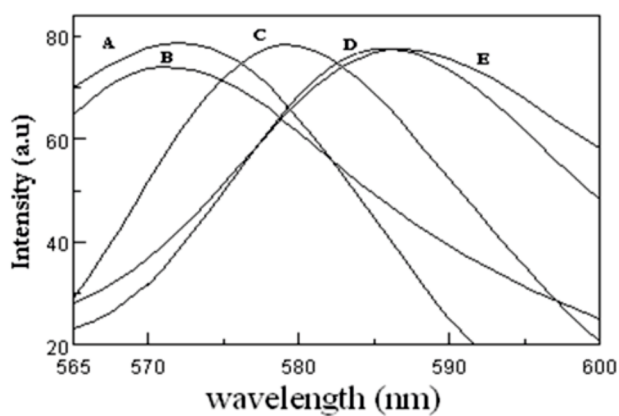


Fig.3: Emission spectrum of RB in SDS (A), Water (B), Brij-35 (C), Ethanol (D), CTAB (E)

Table. 1. Photophysical parameters of the Rose Bengal in different media

Sr. No	Solvent	$\lambda_{abs}$ (nm)	$\lambda_{ex}$ (nm)	$\lambda_{ems}$ (nm)	Stokes' Shift $cm^{-1}$
1	SDS	542	562	572	103
2	CTAB	552	552	587	111.4
3	Brij-36	560	555	579	121
4	Water	540	545	571	82
5	Ethanol	562	569, 530	586	58.4

#### 4. CONCLUSION:

The visible region absorption of Rose Bengal exhibits a relatively small solvatochromic shift in the solvents studied, consistent with the assignment of the transition to  $\pi-\pi^*$ . The solvatochromic shifts for the absorption and fluorescence maxima was nearly 700 nm.

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